www.ijrtem.com Volume 2 Issue 12 | December 2018 | PP 18-27

Pharmacognostic investigation of dried powdered fruits with seeds of the traditional medicinal plant Xylopia aethiopica used for the treatment of both external and internal pains in Sierra Leone

^{1&2},Lahai Koroma, ²,L.M. Kamara, ², T.B.R. Yormah, ³,G.M.T. Robert

1, Department of Basic and Environmental Sciences, Eastern Polytechnic, Kenema, Sierra Leone ^{2,} Department of Chemistry, Fourah Bay College, University of Sierra Leone, Sierra Leone ^{3,} Department of Chemistry, Njala University, Njala, Bo District, Sierra Leone

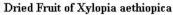
ABSTRACT: Pharmacogenetic investigation of dried powdered fruit with seeds of the traditional medicinal plant Xylopia aethiopica used for the treatment of both internal and external pains in Sierra Leone has been carried out. The results of the investigation indicated colour and taste dried powdered fruit with seeds of X. aethiopica to be light brown in colour, spicy and bitter taste. The powdered plant organ also gave fluorescent derivatives with NaOH solution, ammonia solution, 50% HCl and 50% HNO3 when viewed under UV/Lamp confirming the presence crude drugs in the plant organ investigated. Phytochemical evaluation of the plant organ revealed moderate to high contents of carbohydrates, alkaloid, flavonoids, proteins sterols/terpenes, tannins and phenolic compounds and saponins in the Ethanolic, methanol and aqueous extract. Elemental analysis of the dried powdered fruit with seeds of X. aethiopica was performed with a Niton XL3t GOLD + Hand held X-ray Fluorescence (Thermo Fisher). The Niton Hand held **XRF Instrument** uses Ag-anode X-ray tube with a voltage of 50kV and eauipped with a Si-drift detector (SDD). Accurate energy and efficiency calibrations of the spectrometer were made using a certified reference material - SRM 1573a - Tomato Leaves supplied by the International Energy Agency (IAEA), Vienna, Austria. The spectrum acquisition time was 480sec for the sample and the dead time was around 50%. The results indicated that the plant organ contained large amounts of nutrients and were rich in K (42283 \pm 194.00 ppm), Ca (8682 \pm 80.00 ppm), Mg (4016 \pm 1216 ppm), Al (2600 \pm 196.00 ppm) and Fe (768.22 \pm 14.36 ppm). The other elements present in smaller quantities were Ti (211 \pm 15.00 ppm), **Rb** (62.24 \pm 1.00 ppm), **Sr** (39.87 \pm 0.71 ppm) **Zr** (23.74 \pm 0.72ppm), **Zn** (22.09 \pm 1.96 ppm), **Sc** (22.00 \pm 11.00 ppm), Cu (9.99 \pm 4.03 ppm) and Mo (4.92 \pm 0.73ppm). Only two elements Mn and V were out of limit of detection of the equipment. The above elements have been reported to play great role in metabolic processes in humans thus preventing various types of mineral deficiency diseases that could be associated with pains and degenerative diseases.

KEY WORDS: Pharmacognosy, therapeutic efficacy, phytochemicals, herbal medicine and mineral analysis

INTRODUCTION

This research is geared towards the Pharmacognostic investigation of dried powdered fruit with seeds of the traditional medicinal plant Xylopia aethiopica used for the treatment of both internal and external pains in Sierra Leone. There are a lot of traditional healers in Sierra Leone some of whom just wake up in the morning and start using traditional herbs to cure patients. The therapeutic efficacy of traditional medicinal plants depends upon the quality and quantity of chemical constituents which are unknown to the traditional healers. The misuse of herbal medicine or natural products starts with wrong identification of the plant. The most common error is one common vernacular name given to two or more entirely different species [1]. Unlike taxonomic identification, pharmacogenetic study comprising Phytochemical, Organoleptic, Physicochemical, Fluorescence analysis and mineral analysis of dried powdered fruit with seeds of the traditional medicinal plant Xylopia aethiopica helps in identifying adulteration of the dried powdered form of the plant organ. The powdered fruit with seeds is spicy and you have a lot other spicy plants. Figure 1 below shows the images of the dried fruit, the ripe fruit with part of the branches and leaves and fresh green fruit of Xylopia aethiopica. Figure 2 shows the diagram of the Leaves, Flowers and Fruit bunch and Figure 4 the image of the powdered plant material.







Fresh riped Fruit with Leaves



Fresh green Fruits of Xylopia aethiopica

FIGURE 1

Local vernacular names in Sierra Leone

Creole: SPAIS-TIK Mende: Hewe Temne: Ma-TEL

Kissi: SIAWO Karim: So

Xylopia aethiopica is an evergreen, aromatic tree belonging to the Annonaceae family and can grow up to 20 m high. It grows very well in the Tropical rain forest of Africa. It is found in most African Countries [2], [3], [4], [5], [6], [7] and [8] and grows in riverine and fringing forest, and as a pioneer species in arid savanna regions. [3] In West Africa and in Sierra Leone in particular, harvesting time of the fruit runs from February to May and again from August to October [9] each year. The tree is widely distributed in the forest zones; it is present in the coastal savanna and extends into the interior savanna in forest remnants. It is essentially a tree of secondary forest and often forms pure stands in bush 10-20 years old. The fruits are reported to be smoked like tobacco, and rubbed directly on the head to cure headaches and hot decoction taken to cure constipation.

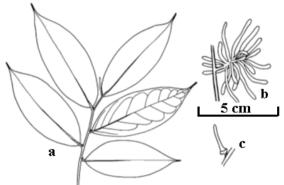


Figure 2: Diagrams of a. Leaves, b. Fruits and c. Flower bud of Xylopia aethiopica

An infusion of the plant's Stem bark or fruit is used in the treatment of bronchitis, dysenteric conditions, or as a mouthwash to treat toothaches, febrile pains, to treat asthma, stomach-aches and rheumatism.[5] It remains an important item of local trade throughout Africa as a spice, and flavouring for food and for medicine. The fruit is sometimes put into jars of water for purification purposes [5]. The dried fruits of *X. aethiopica* (Grains of Selim) are used as a spice and an herbal medicine. In Sierra Leone the sun-dried fruit with seeds are cooked and the soup drunk as a remedy for internal pains and severe stomach aches. The powdered fruit and seeds are also grounded with ginger and used externally as pain reliever. The hot decoction is drunk to induce foetal movement during pregnancy. Trace elements are essential components of biological structures that mediate vital effect on and play a key role in a variety of the biochemical processes necessary for life. Excessive levels higher than that needed for biological functions of these elements can be toxic for the body health. Hence any Pharmacognostic investigation of traditional medicinal plants without mineral analysis cannot be completed.

II. MATERIALS AND METHODS

Collection and preparation of dried plant materials: Fresh ripped fruits of *Xylopia aethiopica* were harvested with the inflorescence from the Gola Rain Forest and sun-dried for 4-7 days. The fruits were not dried on the ground, but on a protective cloth to minimize any microbial contamination.

After drying, the fruits are removed from the inflorescence stalks. It was then reduced in size by crushing it into smaller pieces with cutlass using the hand. After the plant material had been dried, it was grounded using a laboratory mill and kept in a proper container until the time of the extraction. The plant organ investigated is the Fruit (F) with seeds of *Xylopia aethiopica*, the image of plant with leaves and fruits **Figure 1** and diagrams of the Leaves, fruits and the flower bud shown in **Figures 2**. A voucher specimen **No. 401** of Powdered fruit with seeds and the whole leaves of *Xylopia aethiopica* was deposited in the Herbarium of the Botany Department, Fourah Bay College (University of Sierra Leone). The plant material was used to carry out the following analyses described below:

Organoleptic evaluation Fluorescence analysis Phytochemical screening Mineral analysis

III. EXPERIMENTAL

Organoleptic characters: Organoleptic evaluation was carried out by means of sense organs, which provide the simplest as well as quickest means to establish the identity and purity to ensure quality of a particular drug. Organoleptic characters investigated [10] are size, colour, odour, taste and texture of the dried powdered fruit with the seeds of *Xylopia aethiopica*.

The results are shown in **Table 1** and the image of the dried powdered fruit with the seeds of *Xylopia aethiopica* in **Figure 2**

Fluorescence analysis:0.5mg of dried powdered fruit with seeds of *Xylopia aethiopica* was placed in a glass petri dish free from grease and 2-3 drops freshly prepared reagent solution was added, mixed gently with a glass rod and allowed to for few minutes. The following freshly prepared reagents were used; 1 N NaOH (aq), 1 N NaOH (alc.), Ammonia, Picric acid, Petroleum ether, 50% HCl, 50% H₂SO₄, 50% HNO₃, Ethyl acetate, Ethanol, Methanol, and Bromine water. The colours of each of the contents in Petri dish were observed in visible light, short (254 nm) and long (365 nm) ultra violet radiations using a U/V Lamp. A piece of white paper was dipped in each of the solutions and viewed using both visible light and under the U/V Lamp to compare the colours obtained. The colours observed by application of different reagents in different radiations are recorded [11] as shown in Table 2.

Phytochemical analysis: Soxhlet extraction was carried out on the dried powdered fruit with seeds of *Xylopia aethiopica* using solvents of increasing polarity (i.e. Petroleum ether [60-80 °C], Acetone, Chloroform Methanol, 95% Ethanol and Water. Each of the solvent extracts was concentrated, reduced to a semisolid mass using a Rotary Evaporator at 50°C and kept in a special containers for phytochemical screening. The Phytochemical screening involved testing each of the **Solvent Extracts** for the various classes of secondary plant metabolites. The methods used for detection of various phytochemicals were followed by qualitative chemical test and by standard procedures [14, 15] to give general idea regarding the nature of constituents present in each of the solvent extracts of the plant part investigated [16, 17, 18, 19, 20, 21 & 22]. They are generally tested for the presence secondary plant metabolites such as Carbohydrates, alkaloids, tannins/phenolic compounds, flavonoids, Sterols/triterpenes, Amino acids/ proteins and saponins/glycosides etc.

Test for Carbohydrates: A small quantity each of the Solvent Extract was dissolved in 5 ml distilled water and filtered. The filtrates were subjected to the following tests to detect the presence of carbohydrates. Molisch's test:1ml of each of the extract filtrates was treated with 2 drops of alcoholic α -naphthol solution in a test tube and 1 ml of concentrated tetraoxosulphate (VI) acid was added carefully along the sides of the test tubes. Formation of violet/purple ring at the junction may indicate the presence of carbohydrates.

Test for reducing sugars: Fehling's test: - 1ml of each of the **e**xtract filtrate was treated in equal volumes with 1ml Fehling A and 1ml Fehling B solutions, boiled for one minute. The mixtures were boiled for 5-10 minutes on water bath. The formation of Reddish brown precipitate due to formation of cuprous oxide indicates the presence of reducing sugar.

Benedict's test: - 1ml of each of the extract filtrate was treated with equal volumes of Benedict's reagent in test tubes. The mixtures were boiled for 5-10 minutes on water bath. A change in colour of the solution from blue to green, to yellow or brick-red precipitate depending on amount of test item present indicates the presence of reducing sugar.

Iodine Test: 2-3 drops of iodine solution was added to 1ml of each of the solvent extracts. The formation blue-black colour indicates the presence of starch.

Test for Saponins: Froth test: - Each of the **Solvent Extract** was treated with water in a tube shaken well. The appearance of a persistent froth on the top of the mixture indicates the presence of Saponins.

Tests for Amino acids and Proteins: Biuret test (General test):- Each of the **Solvent Extract** was treated with 1 ml 10% sodium hydroxide solution and heated. 2-3 drops of 0.7% copper (II) tetraoxosulphate (VI) solution was added to the mixture stirred and allowed to stand for few minutes. The formation of purplish violet colour may indicate the presence of proteins.

Millions Test (for proteins):-3 ml of each of the **Solvent Extract** was mixed with 5 ml Million's reagent separately. The formation of white precipitate which on heating turned to brick red indicated the presence of amino acids.

Tests for Sterols and Triterpenoids:

Libermann-Burchard test: The each **Solvent Extract** was treated with few drops of acetic anhydride boiled for few minutes. The mixture was cooled and concentrated tetraoxosulphate (VI) acid added down the side of the test tubes. A brown ring at the junction of two layers with the upper layer turning green indicates the presence of sterols while formation of deep red colour indicates the presence of triterpenoids.

Salkowski's test: Each of the **Solvent Extract** was treated with chloroform with few drops of concentrated tetraoxosulphate (VI) acid, shaken well and allowed to stand for some time. The appearance of red colour in the lower layer indicates the presence of sterols while formation of yellow coloured lower layer indicates the presence of triterpenoids.

Tests for tannins and phenolic compounds: Ferric chloride test. Small amount each of the **Solvent Extract** was shaken with water and warmed. 2 ml of 5% ferric chloride solution was added and observed. The formation of green or blue colour indicates the presence of phenols.

Gelatin test: 1% gelatin solution containing 10% sodium chloride was added to each of the **Solvent Extract**. The formation of precipitate indicates the presence of tannins and phenolic compounds.

Test for alkaloids: About 50 mg of each of the **Solvent Extract** was stirred with about 5 ml of dilute hydrochloric acid separately and filtered. Each filtrate was tested with the following reagents:

Dragendroff's test: Few drops of Dragendroff's reagent (solution of potassium bismuth oxonitrate iodide) was added to each filtrate and observed. The formation of orange yellow precipitate indicates the presence of alkaloids.

Mayer's test: Few drops of Mayer's reagent (Potassium mercuric iodide solution) was added to each filtrate and observed. The formation of white or cream colour precipitate indicates the presence of alkaloids.

IV. TESTS FOR FLAVONOIDS:

Shinoda's test (Magnesium Hydrochloride reduction test)

5ml. 95% ethanol was added separately to each of the **Solvent Extract**. Each mixture was treated with 0.5g magnesium turnings and few drops of conc. HCl. The formation of pink colour indicates the presence of Flavonoids.

Lead acetate test: Lead acetate solution was added a small quantity of each of the **Solvent Extract** and observed. The appearance of yellow colour precipitates after few minutes indicates the presence of Flavonoids. Results are shown in **Table 3**

V. MINERAL ANALYSIS

Sample preparation: Sample was thoroughly washed with pure water and rinsed with double distilled water in order to remove the sand or dust particles and all other surface contamination. The plant sample was then air dried, grounded and homogenized in an agate mortar and sieved through a 250µm diameter sieve. A quantity of 3.0g mass of the powdered sample was weighed with an analytical balance and placed in a sample cup holder.

Sample analysis: Elemental analysis of the sample was performed with a Niton **XL3t GOLD** + Hand held X-ray Fluorescence (Thermo Fisher). The Niton Hand held **XRF Instrument** uses Ag-anode X-ray tube with a voltage of 50kV and equipped with a Si-drift detector (SDD). Accurate energy and efficiency calibrations of the spectrometer were made using a certified reference material – SRM 1573a – Tomato Leaves supplied by the International Energy Agency (IAEA), Vienna, Austria. The spectrum acquisition time was 480sec for the sample and the dead time was around 50%.

X-Ray Fluorescence has long been recognized as a powerful technique for the qualitative and quantitative elemental analysis [23, 24]. It has the advantage of being non-destructive, multi-elemental, fast and cost-effective. Furthermore, it offers a fairly uniform detection limit across a large portion of the Periodic Table and is applicable to a wide range of concentrations. In this study, a total of fifteen elements (K, Ca, Mg, Al, Ti, V, Mn, Fe, Cu, Zn, Rb, Sr, Zr, Mo, and Sc) were determined in the powdered fruit with seeds of medicinal plant *Xylopia aethiopica* by using EDXRF. The mean concentrations of various metals in the plant sample are shown in **Table 4**.



Figure 3: EDXRF used for elemental analysis of powdered plant sample

VI. RESULTS AND DISCUSSIONS

Organoleptic evaluation of the dried powdered fruits with seeds of *Xylopia aethiopica* **plant:** The results of organoleptic evaluation of the dried powdered fruits with seeds of *Xylopia aethiopica* plant are reported in **Table1** below with the photo of the dried powdered fruits with seeds of *Xylopia aethiopica* plant shown in **Figure 3**

Table 1: Showing the results of organoleptic evaluation of dried powdered fruits with seeds of *Xylopia* aethiopica plant

PLANT ORGAN	PROPERTY TESTED						
INVESTIGATED	COLOUR	ODOUR	TASTE	TEXTURE	PARTICLE SIZE		
Dried powdered fruits with seeds	Light Brown	spicy	Bitter	Powdered	100 # wire gauge		

The bitter taste indicates that each of the powdered plant materials contain alkaloids. The Light brown colour of the powdered plant material shown in **Figure 4** will also help who so ever wish to buy and use the plant material for medicinal purpose. It helps prevent adulteration.



Figure 4: Powdered Fruit with seeds xylopia aethiopica

Fluorescence analysis of the dried powdered fruits with seeds of *Xylopia aethiopica* plant: The results of Fluorescence analysis carried out on the dried powdered fruits with seeds of *Xylopia aethiopica* plant are reported in **Table 2** below.

TABLE 2: Results of fluorescence analysis

Test	Powdered plant material	Colour in Visible/day light	Colour under Ultra violet light		
1	Powder	Brown	Brown		
2	Powder + 1M NaOH(aq)	Brown	Light orange		
3	Powder + 1M NaOH(alc)	Brown	Bright Orange		
4	Powder + Ammonia	Orange	Bright orange		
5	Powder + Picric acid	Light green	Yellow		
6	Powder + Petroleum ether	Light brown	Black		
7	Powder + 50% HCl	Brown	Light blue		
8	Powder + 50% H ₂ SO ₄	Brown	Brown		
9	Powder + 50% HNO ₃	Brown	Cream white		
10	Powder + ethyl acetate	Brown	Brown		
11	Powder + Ethanol	Light orange	Black		
12	Powder + Methanol	Light orange	Black		
13	$Powder + Br_{2(aq)}$	Light orange	Black		

The above table showed a colour change in reagents like Powder + 1M NaOH(aq), Powder + 1M NaOH(alc.), Powder + Ammonia, Powder + 50% HCl, and Powder + 50% HNO₃. Some constituents show fluorescence in the visible range in daylight. The ultra violet light produces fluorescence in many natural products which do not visibly fluoresce in daylight. If substance themselves are not fluorescent, they may often be converted into fluorescent derivatives or decomposition products by applying different reagents as illustrated above. Fluorescence analysis is one of the parameters for pharmacognostic evaluation of crude drugs [14] in traditional medicinal plants. Thus, the process of standardization can be achieved by stepwise pharmacognostic studies as stated above. This research work helps in identification and authentication of the dried powdered fruits with seeds of *Xylopia aethiopica* plant material used in traditional medicine. Such information can act as reference information for correct identification dried powdered fruits with seeds of *Xylopia aethiopica* plant and also will be useful in making a monograph of the plant. Further, it will act as a tool to detect adulterants and substituent and will help in maintaining the quality, reproducibility and efficacy of natural drugs.

Phytochemical screening carried out on the dried powdered fruits with seeds of *Xylopia aethiopica* plant The results of phytochemical screening carried out on the dried powdered fruits with seeds of *Xylopia aethiopica* plant are shown in **Table 3** below;

TABLE 3: Results of phytochemical screening carried out on the dried powdered fruits with seeds of *Xylopia aethiopica* plant

Experiment		Solvents						
Secondary Plant Metabolites Tests/Reagents		PZ	AC	CHL O	MeO H	EtOH	Water	
	Molisch's Test	-	-	+	++	++	++	
Carbohydrates	Fehling's Test	-	+	+	++	++	++	
	Benedicts Test	-	-	+	++	++	++	
	Iodine Test	-	-	-	++	++	++	
	Mayer's Test	+++	++	+	+++	+++	+++	
Alkaloids	Dragendroff's Test	++	++	+	++	++	++	
Tannins	Iron(III)Chloride Test	-	-	+	-	+	++	
	Gelatin Test	-	++	-	+	++	+	

	Shinoda's Test	-	-	+	++	++	+++
Flavonoids	Lead acetate Test	-	+	+	++	++	+++
Sterols/Triterpene	Libermann-Burchard	-	+	+	++	+++	+++
S	Test						
	Salkowski's Test	-	+	+	++	+++	+++
Amino acids and	Biuret Test	-	-	-	+	+	++
Proteins	Million's Test	-	-	-	+	+	++
Saponins	Froth Test	-	-	+	++	++	+++

KEY: PZ = Petroleum ether, AC = Acetone, CHLO = Chloroform, MeOH = Methanol, EtOH = Ethanol ; + + + = Intense; + + = Moderate; + = Slight; - = Absent

Petroleum ether, acetone, chloroform, methanol, ethanol and aqueous crude extracts of the dried powdered fruits with seeds of *Xylopia aethiopica* plant used for the treatment of both internal and external pains in Sierra Leone was evaluated for the presence of secondary plant metabolites.

The Phytochemical evaluation according to **Table 3**, revealed moderate to high contents of carbohydrates, alkaloid, flavonoids, proteins sterols/terpenes and saponins in the Ethanolic, methanol and aqueous extract. All of the solvent extracts revealed moderate concentration of Tannins and Phenolic Compounds. The petroleum ether and acetone extracts gave the least concentration of the phytoconstituents investigated. The detection of the above secondary plant metabolites supports the use of the plant in traditional medicine

Table 4: Showing the total contents of elements (in ppm) in the powdered fruit with seeds of Xylopia aethiopica

Plant Organ	K	± SD	Ca	± SD	Mg	± SD	Al	± SD
Powdered fruit								
with seeds	42283	194.00	8682	80.00	4016	1216	2600	196.00
Plant Organ								
	Ti	± SD	V	± SD	Mn	± SD	Fe	± SD
Powdered fruit	211	15.00	<1.0D	0.00	< LOD	1.4.40	769.22	14.26
with seeds	211	15.00	< LOD	8.08	< LOD	14.49	768.22	14.36
Plant Organ								
	Cu	± SD	Zn	± SD	Rb	± SD	Sr	± SD
Powdered fruit with seeds	9.99	4.03	22.09	1.96	62.24	1.00	39.87	0.71
with seeds	7.77	4.03	22.07	1.70	02.24	1.00	37.07	0.71
Plant Organ								
C	Zr	± SD	Mo	± SD	Sc	± SD		
Powdered fruit								
with seeds	23.74	0.72	4.92	0.73	22.00	11.00		

$LOD = Limit of detection (Not available) \pm SD = Standard deviation$

The results of the current study as shown in **Table 4** revealed that all the metals investigated (**K**, **Ca**, **Mg**, **Al**, **Ti**, **V**, **Mn**, **Fe**, **Cu**, **Zn**, **Rb**, **Sr**, **Zr**, **Mo**, **and Sc**) were accumulated in greater or lesser extent in the powdered fruit with seeds of *Xylopia aethiopica* plant. The plant organ contained large amounts of nutrients and were rich in **K** (42283 \pm 194.00 ppm), **Ca** (8682 \pm 80.00 ppm), **Mg** (4016 \pm 1216 ppm), **Al** (2600 \pm 196.00 ppm) and **Fe** (768.22 \pm 14.36 ppm). The other elements present in smaller quantities were **Ti** (211 \pm 15.00 ppm), **Rb** (62.24 \pm 1.00 ppm), **Sr** (39.87 \pm 0.71 ppm) **Zr** (23.74 \pm 0.72ppm), **Zn** (22.09 \pm 1.96 ppm), **Sc** (22.00 \pm 11.00 ppm), **Cu** (9.99 \pm 4.03 ppm) and **Mo** (4.92 \pm 0.73ppm). The other two elements **Mn** and **V** were out of limit of detection of the equipment. Below are some of the uses of the above elements in various metabolic processes in human beings and animals.

Potassium participates actively in the maintenance of the cardiac rhythm [25] and in constipation. Ca is the main constituent of the skeleton and is important for regulating many vital cellular activities such as nerve and muscle function, hormonal actions, blood clotting and cellular mortality. Iron is an essential element for human beings and animals and is an essential component of hemoglobin. It facilitates the oxidation of carbohydrates, protein and fat to control body weight, which is very important factor in diabetes. Zinc is the component of more than 270 enzymes [26] and its deficiency in the organism is accompanied by multisystem dysfunction. Zn has been reported to be responsible for sperm manufacture, fetus development and proper function of immune response [27] in humans. Zinc makes a very specific contribution in the breakdown of carbohydrate and is involved in the granulation and storage of insulin in the beta cells of the pancreas [28]. It has been shown that the defect of dysinsulinemia, a pre-diabetic state, is caused by the inability of the beta cells to store and granulate insulin [29]. Zn deficiency causes hair loss, delayed sexual maturation, impotence, hypogonadism in males, and eye and skin lesions, weight loss, delayed healing of wounds, taste abnormalities, and mental lethargy can also occur [30-32]. The ability of trace elements to function as substantial affecter in a variety of the processes necessary for life, such as regulating homeostasis and prevention of free radical damage, can provide an answer to the definite correlation between content of trace elements and many common diseases.

Copper is an essential nutrient that plays an important role in the production of hemoglobin, myelin, collagen and melanin [33]. Magnesium also plays a very important role as a catalyst in several hundreds of biological reactions, mostly in the glycolytic enzymes [34]. Studies have indicated that inadequate Mg intake frequently causes muscle spasms and has been associated with cardiovascular disease, diabetes, high blood pressure, anxiety disorders, migraines, osteoporosis, and cerebral infarction [35, 36-39] which support the use of the plant as remedy for pains. It has been reported that medicinal plants possessed some important elements which have both therapeutic and prophylactic properties [40-44]. Excessive levels of these elements in medicinal plants could lead to toxicity. Hence knowledge of the presence and amount of these elements in plants also validates the use of the plant as food, medicine and as an important parameter during pharmacognostic analysis. Fruits and vegetables are reported to be a safe and valuable source of minerals [41].

VII. CONCLUSION

Organoleptic evaluation, fluorescence analysis, phytochemical screening and mineral analysis were carried out on the dried powdered Leaves of *X. aethiopica* plant used for the treatment of both internal and external pains in Sierra Leone. The results of organoleptic evaluation, fluorescence analysis and phytochemical screening indicate the presence of crude pharmaceuticals that support the use of the plant in traditional medicine. The result of mineral/elemental analysis of the plant indicated that the plant organ investigated contained large amounts of nutrients and was rich in K (42283 \pm 194.00 ppm), Ca (8682 \pm 80.00 ppm), Mg (4016 \pm 1216 ppm), Al (2600 \pm 196.00 ppm) and Fe (768.22 \pm 14.36 ppm).

The other elements present in smaller quantities were **Ti** (211 \pm 15.00 ppm), **Rb** (62.24 \pm 1.00 ppm), **Sr** (39.87 \pm 0.71 ppm) **Zr** (23.74 \pm 0.72 ppm), **Zn** (22.09 \pm 1.96 ppm), **Sc** (22.00 \pm 11.00 ppm), **Cu** (9.99 \pm 4.03 ppm) and **Mo** (4.92 \pm 0.73 ppm). The other two elements **Mn** and **V** were out of limit of detection of the equipment.

The human body plays a key role in regulating the amount of trace metals circulating in blood and stored in cells. **Zn** is the second most abundant metals in organisms after iron and appears in all enzyme classes, while copper is present in every tissue of the body, but is stored primarily in the liver, with fewer amounts found in the brain, heart, kidney, and muscles. Copper plays an important role in our metabolism, as it allows many critical enzymes to function properly. Copper is essential for maintaining the strength of the skin, blood vessels, epithelial and connective tissue throughout the body. Cu plays a role in the production of hemoglobin, myelin, melanin and it also keeps thyroid gland functioning normally. Iron is involved in the binding, transporting, and release of oxygen in higher animals. Iron, zinc, and selenium are essential components of enzymes where they attract or subtract molecules and facilitate their conversion to specific end products. Some of the trace elements control important biological processes by facilitating the binding of molecules to their receptor sites on cell. The above elements have been reported to play great role in metabolic processes in humans thus preventing various types of mineral deficiency diseases that could be associated with pains and degenerative diseases.

VIII. RECOMMENDATIONS

This research work has shown that the colour and taste dried powdered fruit with seeds of *X. aethiopica* to be light brown in colour, spicy and bitter taste. Phytochemical evaluation of the plant organ revealed moderate to high contents of carbohydrates, alkaloid, flavonoids, proteins sterols/terpenes, tannins and phenolic compounds and saponins in the Ethanolic, methanol and aqueous extract. Mineral/elemental analysis of the plant indicated that the plant organ contained large quantities of *K*, *Ca*, *Mg*, *Al*, and *Fe*.

- **Ti, Rb, Sr, Zr, Zn, Sc, Cu** and **Mo** were also present in smaller quantities. The detection of secondary plant metabolites and above elements supports the use of the plant in traditional medicine and provides pertinent information for traditional healers who wish to trade with the powdered fruit with seeds of *X. aethiopica*. In view of this it is therefore recommended that:
- Traditional healers must use this information to determine the quality and quantity of traditional herbs that should be given to patients since intake of excess of the trace elements can be toxic..
- Patients are able to identify the colour and taste of the dried powdered fruit with seeds of *X. aethiopica* plant in order to avoid adulteration
- Further work to be done in order to isolate and characterize secondary plant metabolites from the plant that provides remedy for pains and other degenerative diseases..

ACKNOWLEDGEMENT

The authors are grateful to Mr. Anthony J. Kamara, Department of Physics for carrying out elemental analysis of the dried powdered fruit with seeds of *Xylopia aethiopica* plant using EDXRF, Laboratory technicians of the Department of Chemistry, Fourah Bay College, University of Sierra Leone and the Principal Eastern Polytechnic, Kenema for providing financial assistance

REFERENCES

- [1] **Dinesh Kumar C**. (2007) Pharmacognosy can help minimize accidental misuse of herbal medicine. Curr. Sci 2007; 3:1356-1358.
- [2] **A. Rich.** (2008) Taxonomy for Plants. <u>USDA</u>, <u>ARS</u>, National Genetic Resources Program, National Germplasm Resources Laboratory, Beltsville, Maryland. *Retrieved* 2008-04-19.
- [3] *Orwa; et al.* (2009). "*Xylopia aethiopica*" (*PDF*) Agroforestry Database 4.0. World Agroforestry Center. Retrieved 1 January 2013.
- [4] **A. Rich.** (2013) African Plant Database. <u>Conservatoire et Jardin botaniques de la Ville de Genève</u>. 16 April 2007. Retrieved 1 January 2013.
- [5] Harris, D.J., Moutsamboté J.-M., Kami, E., Florence, J., Bridgewater, S. & Wortley, A.H. (2011).

 "An introduction to the trees from the North of the Republic Congo". Royal Botanic Garden Edinburgh.

 Retrieved 1 January 2013.
- [6] **Burkill, Humphrey Morrison** (1985). Entry for Xylopia aethiopica Dunal A. Rich.: family ANNONACEAE. The useful plants of west tropical Africa, Vol 1. JSTOR. Retrieved 1 January 2013.
- [7] **Mitani, M. (1999).** Does fruiting phenology vary with fruit syndrome? An investigation on animal-dispersed tree species in an evergreen forest in south-western Cameroon. Ecological Research, 1999 14:371-383.
- [8] **A. Rich**. (2013) <u>Missouri Botanical Garden</u>. "<u>Distributions:Xylopia aethiopica (Dunal)</u>". Tropicos.org. Retrieved 1 January 2013.
- [9] **Burkill HM.** (1995). The useful plants of West Tropical Africa. vol. 3. Caesalpinioideae, pp. 50-177. Royal Botanic Gardens, Kew. 857 pp. 1995
- [10] **Siddiqui, Hakim MA**. **(1995)** Format for the pharmacopoeia analytical standards of compound formulation, workshop on standardization of Unani drugs, (appendix), 24-25 January. New Delhi: Central Council for Research in Unani Medicine (CCRUM); 1995.
- [11] Kokoski J, Kokoski R, Salma FJ. (1958) Fluorescence of powdered vegetable drugs under ultraviolet radiation. J Am Pharm Ass 1958; 47:715-717
- [12] **Tatiya A, Surana S, Bhavsar S, Patil D, Patil Y.** (2012) Pharmacognostic and preliminary phytochemical investigation of Eulophia herbacea Lindl. Tubers (Orchidaceae). Asian Pac J Trop Disease 2012; 2(Suppl 1):S50-55.
- [13] Harborne JB. (1973) Phytochemical methods. Edn 2. London: Chapman & Hall, 1973.
- [14] Kokate CK. (1997) Practical Pharmacognosy, Edn 4, Vallabh Prakashan, Delhi, 107-111, 1997.
- [15] **Zhao Z, Liang Z, Guo P. (2011)** Macroscopic identification of Chinese medicinal materials: Traditional experiences and modern understanding. J Ethnopharmacol 2011; 131:556-561.
- [16] Khandelwal KR: (1995) Practical Pharmacognosy, Nirali Prakashan, 1995, 149-155.
- [17] **Trease E.G. and Evans W.C.** (1978) Pharmacognosy 1978, 11th Edition, Balliere Tindall, London 115-222.
- [18] **Sazada S, Arti V, Ayaz A, Faraha J, Maheswari MK**: (2009) Preliminary Phytochemical analysis of Some Medicinal and Aromatic Plants. Adv. In Biological Res., 2009; 3(5-6): 188-5.
- [19] **Kokate C.K., Purohit A.P. and Gokhale S.B.(2006)** Pharmacognosy. 34th Ed: 2006 Nirali Prakashan, Pune, India.

- [20] Nayak BS, Isitor G, Davir EM and Pillai GK. (2007). The evidence based Wound Healing Activity of Lawsonia inermis Linn. Phytotherapy Research 2007; 29: 829.
- [21] **Sofowora A (1993)**. Medicinal Plants and Traditional Medicine in Africa (2nd ed.) Spectrum Books Ltd. Ibadan, pp 255-256.
- [22] Trease GE, Evans WC (2002). Pharmacognosy (13th ed.). Bailliere Tindall, London, pp. 214-393.
- [23] Queralt I, Ovejero M, Carvalho ML, Marques AF, Liabres JM. (2005) Quantitative determination of essential and trace element content of medicinal plants and their infusions by XRF and ICP techniques. X Ray Spectrom 2005; 34: 213-217.
- [24] Shendkar CD, Chandrachood PS, Pawar AB, Lavate SM, deshpande NR. (2011) Quantitative estimation of macro, micronutrients and trace elements by X-ray fluorescence spectroscopy (XRF) from Achyranthes aspera Linn. Int J Chem Tech Res 2011; 3(2): 610-613.
- [25] Martin Jr, D. W., Mayers, P. A., Rodwell, V. W., Granner, D. K. (1985). Harper's Review of Biochemistry, 20th ed., Lange Medical Publications, California, pp. 651-660. 1985
- [26] **Zinpro Corporation.** (2000). Epithelial tissue: body's first line of defense depends upon trace minerals. Trace Miner Focus. 6: 1-8, 2000
- [27] Serfor-Armah, Y., Nyarko, B. J. B., Akaho, E. H. K., Kyere, A. W. K., Osae, S. and Oppong-Boachie, K. (2002). Multielemental analysis of some traditional plant medicines used in Ghana. J. Trace Microprobe Tech. 20: 419-427. 2002
- [28] **Bollin Se et al.(1964)** The structure and Metabolism of the Pancreatic Islets. Oxford, Pergamon Press, 1964.
- [29] **Zilva JF, Pannal PR:** (1972) Clinical Chemistry in Diagnosis and Treatmeat, Chicago, Yearbook Medical Publisher, Inc., 1972.
- [30] C. A. Heyneman, (1996) Zinc deficiency and taste disorders, Annals of Pharmacotherapy, 30: 186-187.
- [31] W. Maretm, H. H. Sandstead, (2006) Zinc requirements and the risks and benefits of zinc supplementation, *Journal of Trace Elements in Medicine and Biology*, 20: 3-18.
- [32] **S. Prasad, (2004)** Zinc deficiency: İts characterization and treatment, *Metal İons in Biological Systems*, **41:** 103-137.
- [33] Cobanoglu, U., Demir, H., Sayir, F., Duran, M. and Mergan, D. (2010) Some mineral, trace element and heavy metal concentrations in lung cancer. Asian Pacific J. Cancer Prev. 11: 1383-1388.
- [34] Guthrie HA: Introductory Nutrition, 3rd Ed. Saint Louis, The C.V. Moshby Co. 1975:1975.
- [35] M. P. Romani, (2013) Magnesium in health and disease. In A. Sigel, H. Sigel, R. K. O. Sigel, (Ed.), *Interrelations between Essential Metal Ions and Human Diseases, Metal Ions in Life Sciences*, Vol. 13. Ch. 3. Dordrecht: Springer, p49-79.
- [36] . S. C. Larsson, M. J. Virtanen, M. Mars, S. Männistö, P. Pietinen, D. Albanes, J. Virtamo, (2008) Magnesium, calcium, potassium, and sodium intakes and risk of stroke in male smokers, Archives of Internal Medicine, 168(5): 2008; 459-465.
- [37] S. M. Kobrin, S. Goldfarb, (1990) Magnesium deficiency, Seminars in Nephrology, 10(6): 525-535.
- [38] H. Geiger, C. Wanner, (2012) Magnesium in disease, Clinical Kidney Journal, 5: 25-38.
- [39] **D. P. Chaudhary, R. Sharma, D. D. Bansal, (2010)** Implications of magnesium deficiency in Type 2 diabetes: A review, *Biological Trace Element Research*, **134:** 119-129.
- [40] Mark PE, Michael JB, Jianwei (2000) WH. Plants as source of concentrated mineral nutritional supplements. Food Chem 2000; 77: 181-188.
- [41] **Haq F, Ullah E. (2011)** Comparative determination of trace elements from Allium sativum, Rheum australe and Terminalia chebula by atomic absorption spectroscopy. IJB 2011; 1(5): 77-82.
- [42] **Harrington KC, Thatcher A, Kemp PD. (2006)** Mineral composition and nutritive value of some common pasture weeds. N Z Plant Prot 2006; 59: 261-265.
- [43] **Haq F, Rehman A, Ahmad H, Iqbal Z, Ullah R.** (2012) Elemental analysis of Paeonia emodi and Punica granatum by atomic absorption spectroscopy. Am J Biochem 2012; 2(4): 47-50.
- [44] **Pandey MAB, Abidi S, Singh RP.** (2006) Nutritional evaluation of leafy vegetables. Paratha. J Hum Ecol 2006; 19: 155-156.